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(54) Title: FACTOR VII OR VIIa-LIKE MOLECULES

(57) Abstract: The present invention relates to novel factor VII (FVII) or Factor VIIa (FVIIa) polypeptide conjugates, to their preparation and use in therapy, in particular for the treatment of a variety of coagulation-related disorders. These novel polypeptide conjugates comprise at least one non-polypeptide moiety covalently attached to a polypeptide, wherein the amino acid sequence of the polypeptide differs from that of wild-type FVII or FVIIa in that at least one amino acid residue comprising an attachment group for said non-polypeptide moiety has been introduced or removed. The conjugates of the present invention have one or more improved properties as compared to commercially available rFVIIa, including increased functional *in vivo* half-life and/or increased plasma half-life, and/or increased bioavailability and/or reduced sensitivity to proteolytic degradation.



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INTERNATIONAL SEARCH REPORT

International application No.

PCT/DK 01/00094

A. CLASSIFICATION OF SUBJECT MATTER		
IPC7: A61K 47/48, C12N 9/64, C07K 14/745, A61K 38/36 According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED		
Minimum documentation searched (classification system followed by classification symbols)		
IPC7: A61K, C12N, C07K		
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched		
Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 9835026 A1 (NOVO NORDISK A/S), 13 August 1998 (13.08.98), page 3, lines 3-11; page 4, line 10 - page 5, line 3; page 6, lines 4-7; page 12, lines 3-36; page 16 --	1-73
X	US 4904584 A (GRAY SHAW), 27 February 1990 (27.02.90), column 2, line 35; column 2, line 6 - column 3, line 3 --	1-14, 28-33, 56-73
X	EP 0370205 A2 (KYOWA HAKKO KOGYO CO., LTD.), 30 May 1990 (30.05.90), page 4, lines 22-33, 44-47; page 6, line 55 - page 7, line 7 --	1-2, 34-73
<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C. <input checked="" type="checkbox"/> See patent family annex.		
* Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family		
Date of the actual completion of the international search		Date of mailing of the international search report
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Name and mailing address of the International Searching Authority European Patent Office P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk Tel(+31-70)340-2040, Tx 31 651 epo nl, Fax(+31-70)340-3016		Authorized officer CAROLINA PALMCRANTZ/EÖ Telephone No.

INTERNATIONAL SEARCH REPORT

International application No.

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C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5580560 A (ELSE M. NICOLAISEN ET AL), 3 December 1996 (03.12.96), the claims	58-62
A	--	64-73
A	WO 9903887 A1 (BOLDER BIOTECHNOLOGY, INC.), 28 January 1999 (28.01.99)	1-2,15-18
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INTERNATIONAL SEARCH REPORT

International application No.
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Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.: **70-73**
because they relate to subject matter not required to be searched by this Authority, namely:
Claims 70-73 relate to methods of treatment of the human or animal body by therapy (PCT, Rule 39.1(iv)): Nevertheless, a search has been executed for these claims. The search has been based on the alleged effects of the compound/composition.
2. ☒ Claims Nos.: **58-62**
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
see next sheet
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see next sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☒ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
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Invention 1, claims 1-2 (partly), 3-14, 28-33 (partly), 56-73 (partly), relates to a conjugate comprising a modified Factor VII polypeptide covalently attached to at least one non-polypeptide moiety, wherein the amino acid sequence of the Factor VII polypeptide differs from that of wild-type Factor VII (defined in SED ID No:1) in at least one introduced or at least one removed **lysine residue**.

Invention 2, claims 1-2 (partly), 15-18, 28-33 (partly), 56-73 (partly), relates to a conjugate comprising a modified Factor VII polypeptide covalently attached to at least one non-polypeptide moiety, wherein the amino acid sequence of the Factor VII polypeptide differs from that of wild-type Factor VII (defined in SED ID No:1) in at least one introduced **cysteine residue**.

Invention 3, claims 1-2 (partly), 19-27, 28-33 (partly), 56-73 (partly), relates to a conjugate comprising a modified Factor VII polypeptide covalently attached to at least one non-polypeptide moiety, wherein the amino acid sequence of the Factor VII polypeptide differs from that of wild-type Factor VII (defined in SED ID No:1) in at least one introduced or at least one removed **aspartic acid or glutamic acid residue**.

Invention 4, claims 1 (partly), 28-32 (partly), 34-55, 56-73 (partly), relates to a conjugate comprising a modified Factor VII polypeptide covalently attached to at least one sugar moiety, wherein the amino acid sequence of the Factor VII polypeptide differs from that of wild-type Factor VII (defined in SED ID No:1) in at least one introduced or at least one removed **attachment group for the sugar moiety**.

However, the international search covers all inventions.

Box I.2

Present claims 58-62 (which independently disclose the modified Factor VII molecule i.e. these claims are not restricted to conjugates) relate to an extremely large number of possible modifications of the wild-type Factor VII molecule. Support within the meaning of Article 6 PCT and disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the compounds claimed. In the present case the claims so lack support, and the application so lacks disclosure that a meaningful search over the whole of the claimed scope is impossible. Consequently, the search has been carried out for modified Factor VII molecules in general and for those parts of the claims which appear to be supported and disclosed, namely those parts related to the modifications T106N, K143+N145T, V253N, R290N+A292T, G291N, R315N+V317T, and K143N+N145T+R315N+V317T (c.f. Example 3).

Box II

Unity of invention exists only when there is a technical relationship among the claimed inventions involving one or more of the same or corresponding "special technical features", i.e. features that define a contribution which each of the inventions makes over the prior art (PCT Rule 13.2).

Initially, all the subject matters were included in the search. The unifying technical feature was considered to be a Factor VII or Factor VIIa conjugate wherein the Factor VII molecule had been modified by introducing or removing at least one amino acid residue comprising an attachment group for the non-polypeptide moiety. However, it soon became obvious that pertinent prior art exists making it necessary to reconsider the technical relationship between the different solutions revealed in the claims. This prior art being WO 9835026 and US 4,904,584.

WO 9835026 relates to polypeptide-polymer conjugates having added and/or removed attachment groups for the polymeric molecule. The attachment groups are e.g. amino groups of lysine, carboxylic acid groups of aspartate and glutamate, and -SH groups (see page 4, line 32-page 5, line 3 and page 12, lines 20-21). The conjugated polypeptide has reduced immunogenicity. It is also stated in the document that even though the polypeptide part of the conjugate can be quite different the principle of the invention may be tailored to the specific type of parent polypeptide (see page 6, lines 4-7).

.../...

US 4,904,584 concerns the modification of biologically active and therapeutically useful polypeptides with a variety of compounds having amine reactive groups e.g. polyethylene glycol (PEG) (see column 1, lines 15-21). The problems with random attachment of the polymer to the amino terminus of the polypeptide and/or at one or more lysine residues in the amino acid sequence of the protein (which may also alter the biological activity of the polypeptide, see column 1, lines 32-46) are solved by site-specific attachment, that is, by modifying the number and position of attachment sites, thus retaining the desirable characteristics of the natural polypeptides (see column 1, lines 46-53). The proteins or polypeptides are modified in amino acid structure relative to the naturally occurring in one or both of the following respects:

- a) at least one lysine residue of the natural compound is deleted or replaced with a substitute amino acid, preferably arginine;
- b) at least one lysine residue is inserted into the natural sequence and/or is used to replace a different amino acid within that sequence (see column 2, line 59-column 3, line 2).

It has been stated in the document that any polypeptide is a candidate for the method of the invention and Factor VII is listed among the polypeptides of interest (see column 2, line 31).

Therefore, in view of the prior art it is considered to be obvious to a person skilled in the art to prepare a conjugate between Factor VII and e.g. PEG, wherein the Factor VII polypeptide differs from that of wild-type Factor VII in at least one introduced or at least one removed attachment site, e.g. a lysine, for the polymeric molecule.

Thus, Factor VII conjugates wherein the Factor VII molecule has been modified by introducing or removing at least one amino acid residue comprising an attachment site for the non-polypeptide moiety are not considered to involve an inventive step in relation to the prior art.

A search for a new common concept of invention, between the different types of Factor VII modifications among the claims, has failed and many of the independent claims represent independent subject matter forming lack of unity with each other. The International Searching Authority has arrived at the following principle of division:

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JA 329084

INTERNATIONAL SEARCH REPORT

Information on patent family members

28/05/01

International application No.

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Patent document cited in search report			Publication date	Patent family member(s)			Publication date
WO	9835026	A1	13/08/98	AU	5749598 A		26/08/98
				CN	1246891 T		08/03/00
				EP	1017794 A		12/07/00

US	4904584	A	27/02/90	AU	2911189 A		19/07/89
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				WO	8905824 A		29/06/89

EP	0370205	A2	30/05/90	AT	168699 T		15/08/98
				DE	68928751 D,T		15/04/99
				ES	2121734 T		16/12/98
				JP	2227075 A		10/09/90
				JP	2928287 B		03/08/99
				US	5218092 A		08/06/93

US	5580560	A	03/12/96	NONE			

WO	9903887	A1	28/01/99	AU	8300098 A		10/02/99
				CN	1269805 T		11/10/00
				EP	1012184 A		28/06/00